THE EFFECT OF DISTILLED WATER UPON THE FIDDLER CRAB.[1]

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At present, *Fundulus* offers the only exception[2] to the statement that the unprotected epithelial surfaces of marine and fresh-water animals are permeable to distilled water and to electrolytes in solution. In all cases life appears to be preserved by some sort of an "adaptation" as yet unexplained which limits the permeability or controls it so as to maintain the integrity of the body fluids. Sumner[3] has shown that the gills in a variety of Teleosts are permeable, and Scott and White[4] have conclusively shown the same thing in *Mustelus*, while Garrey[5] and others have demonstrated the fact in a great number of invertebrates. The apparent exception offered by *Fundulus* is therefore the more remarkable.

While working at the Beaufort Laboratory of the U. S. Fisheries Bureau my friend Professor W. E. Garrey called my attention to the fact that the fiddler crabs (*Uca pugillator*) which abound in that region were passing indifferently from the sea to the overflow of an artesian well and he suggested that this form, like *Fundulus*, might offer an exception to the statement just made regarding the permeability of membranes. A few tests seemed to confirm this, for the crabs appeared to live indefinitely in pure distilled water as well as in salt solutions of various osmotic densities. The writer therefore devoted the greater part of the past summer to a study of the reactions of this animal to various salt solutions. For the privilege of working at the Beaufort Laboratory he is indebted to the kindness of the Hon. George M. Bowers, U. S. Commissioner of Fisheries, and to Mr. Lewis Radcliffe, the

[1] From the Laboratory of the U. S. Bureau of Fisheries at Beaufort and the Zoölogical Laboratory of Washington University.

[2] Species of *Cyprinodon* and *Gambusia* apparently behave in the same way, although the facts have not been thoroughly worked out.

[3] U. S. B. F. Bull., 1905, Vol. 25, p. 97.

[4] *Science*, N. S., 1910, Vol. 32, p. 767.

[5] *Biol. Bull.*, 1905, Vol. 8, p. 257.

director of the laboratory, he is under especial obligation for the constant courtesy and generous assistance of which he was at all times the recipient. The present paper deals only with the effect of pure distilled water upon the fiddler crab.

The crabs inhabit burrows in the sandy or marshy shores, the openings of which are usually, though not always, covered by the tide. At high tide the crabs retreat to their holes which they plug up with sand. As the tide ebbs they emerge and begin feeding, frequently going a considerable distance from their holes. In the interval between high tides, if the sun is bright and the day hot the males go through characteristic and energetic "dances." Altogether they spend many hours of vigorous activity out of the water. A related genus of the same family, *Ocypoda*, lives out of the water almost entirely and spends most of its time running about on the dry sand.[1] In accordance with this habit a peculiar modification has developed in connection with the gill chambers. In these two genera these are very large and cavernous, the two chambers together equalling approximately the space occupied by the viscera. The gills lie in the floor and the large space above them is normally filled with liquid. Communicating with this chamber, in addition to the ordinary openings for the exit and entrance of water, there is an aperture between the basal joints of the third and fourth legs, bounded by chitinized lips which bear tufts of hairs. This opening communicates between the cavity above the gills and the exterior by a sort of canal provided with a ciliated valve. By means of this arrangement, not only is the animal able to store up a large amount of water above the gills but it is also possible for a constant gaseous exchange to take place between this contained water and the air without. In other words the crab when out of the water is able to breathe air.

This accounts for the apparent tolerance of the animal to pure distilled water. When immersed in distilled water it feeds out through this channel some of the sea water contained in its gill chambers. As with some other crustacea a very small amount of NaCl in the surrounding medium is sufficient to preserve the integrity of the membranes and permit the animal

[1] Cf. Cowles. Carnegie Inst. Papers, Tortugas Lab., II., 1908.

to live in spite of the greatly altered osmotic pressure. That the crab accommodates itself in this way was shown by the following experiment. Five crabs that had been running about in a dry tank for a week were rinsed off in running fresh water, dried, and immersed in 250 c.c. of pure distilled water which was removed and renewed at short intervals. These samples were then titrated for chlorides by the Volhard method with the following results:[1]

Interval of Immersion in Pure Distilled Water.	Cl in Milligrams.
30 minutes	0.038
1 hour	0.188
2 hours	0.263
3 hours	0.132
3½ hours	0.297
Total Cl	0.918

The crabs were moribund in another half hour. The gill chamber capacity of each crab was roughly ½ c.c. The percentage of Cl in Beaufort sea water is 1.963 gm. per 100 or .049 gm. to 2½ c.c. The detectible salinity emitted by the crabs was thus only about 20 per cent. of that of sea water. Some may have been "held back," but it is more probable not only that the crabs in the experiment, which had been out of water for a week, but also crabs running about out of the water normally have their gills bathed by a liquid of only about one fifth the concentration of sea water. Experiment has shown that they will live indefinitely when immersed in very much greater dilutions, although I have not found that they will endure such a dilution as that used by Bullot[2] for fresh-water *Gammarus* (0.00008*N*).

In subsequent experiments the side of the gill chamber was cut away and the contents rinsed out with distilled water so that not only were all salts washed away but the gills were directly exposed to the action of the medium. Even under such circumstances the crab does not at once die when placed in distilled water but will live actively usually from four to six

[1] For assistance in carrying out this portion of the experiments as well as for many helpful suggestions the writer is indebted to Mr. Wm. J. Crozier.

[2] Univ. of Calif. Pub. Physiol., 1904, Vol. I., p. 199.

hours. Death appears to be due to a loss of necessary salts by diffusion out of the body fluids together with a disturbance of vital equilibrium on account of the absorption of water. In most marine invertebrates death very quickly follows a transfer into fresh water, that is, the limiting membranes are absolutely permeable to water and osmotic diffusion is very rapid. (The body fluids of all marine invertebrates, so far as known, are isosmotic with sea water.) In *Uca*, on the other hand, the gill membranes are apparently impermeable to water until the strongly solvent power of the distilled water has attacked or partially dissoved them and thus rendered them partially permeable. That they are eventually rendered permeable is shown in the following experiments. A larger species of *Uca* (*U. minax*) was chosen because of its greater size, but the habits and reactions of the two species are identical. The carapace of each crab was cut away on both sides to expose the gills, and the gill chamber was rinsed out. The carapace was then thoroughly dried with absorbent cotton and the crab was carefully weighed, and immersed in 200 c.c. of water distilled in Jena glass over potassium dichromate and sulphuric acid. At intervals this water was changed for a fresh amount and the sample titrated for chlorides with potassium thiocyanate against silver nitrate, using iron alum as an indicator. The following results were obtained:

TABLE I.

I.	II.	III.	IV.	V.	Chlorides (in Mg.) After
0.205					1 hour.
	0.135		7.420	5.312	3 hours.
5.600	9.431	26.09			5 hours.
9.210		1.700	(died)	0.954	7 hours.
					9 hours.
		8.91			11 hours.
15.016	9.516	36.705	(7.42)	6.266	Total chlorides.
7.588	3.544	8.463	6.011	4.638	Original wt. in grams.
7.813		8.460	6.599	4.833	Final weight.
0.225	0.677	0.005	0.588	0.195	Gain in wt.

REMARKS.

Crab No. 1 died shortly after 7 hours.

Crab No. 2 died after 4 hrs., weighed immediately. Big claw cast at beginning, wound seared with nitric acid.

Crab No. 4 died shortly after 7th hr.; final chloride determination not taken.
Crab No. 5 died shortly after 7th hour.

It will be seen that without exception all the crabs gained in weight through immersion in distilled water. Likewise all the crabs lost chlorine, presumably through diffusion. There is no doubt that some of this loss may have been occasioned by bleeding, yet the outer shell of the gill chamber is very little vascular and such a loss must be inconsiderable. It, however, might be sufficient to account for the irregularities in the amounts of chlorine lost by the various crabs. An exception must be made of Crab No. 3 of which the branchiostegite was cut away very deeply and bled into the water. This accounts for the very small gain in weight and exceptionally high chlorine loss. Yet this individual for some reason not evident lived longer than any of the others.

If death is brought about by the extraction of necessary salts from the organism it ought to make considerable difference whether the amount of water that surrounds the animal is great or small. For we have seen that the presence of a very small amount of salt in the water (such as exists in ordinary well water) is sufficient to preserve the membrane or at least to greatly retard its solution by the water. If the amount of water were scant the crabs might be able, by losing a small amount of salts to it and thus raising the osmotic density of the medium ever so slightly, yet thereby to retard the solvent effect of the distilled water on the gill membranes. To test this, fifty tall, wide mouth bottles were arranged, in each ten of which was put, respectively, 50 c.c., 100 c.c., 150 c.c., 200 c.c., and 250 c.c. of distilled water. In each bottle was placed a crab (*U. pugillator*). The gill chambers were not opened but the lot were subjected to running fresh water for fifteen minutes, a procedure which experience has shown displaces practically all the salt water that may be retained in the gill chamber. The results are tabulated below, the figures representing the number of crabs alive at the different intervals of time.

TABLE II.

	50 c.c.	100 c.c.	150 c.c.	200 c.c.	250 c.c.
4 hours	10	10	10	10	10
8 hours	10	10	9	9	8
12 hours	10	10	8	6	2
18 hours	10	7	5	4	0
22 hours	10	1	1	0	
24 hours	9	1	0		
48 hours	6	0			
72 hours	2				

SUMMARY.

1. The fiddler crabs of the genus *Uca* are able to endure abrupt and profound changes in the osmotic density of the water which they enter, passing from sea water into ordinary "fresh" water with impunity.

2. They are able to spend long periods out of the water by the aid of a mechanism by means of which they store up a quantity of water above the gills and aerate it directly by contact with the air.

3. Their gill membranes are only relatively not absolutely impermeable. In the presence of pure distilled water they both lose salts (as indicated by the titratable chlorine) and also gain weight through the absorption of water.

4. They will live a much longer time in small quantities of distilled water than in larger quantities. This is probably due to the fact that the salts diffused out from the tissues although small in amount are sufficient, in a small quantity of water, to raise the osmotic tension of the latter sufficiently to inhibit its solvent action on the gill membranes and thus to retard the further diffusion of salts. In a larger quantity of water this point cannot be reached before the debilitating effect of the loss of salts results in the death of the crabs.